

Patent Claims

1. Method for the proof, quantification and/or
characterization of an analyte (10) contained in a first
5 fluid consisting of the following steps:

a) Bringing into contact and incubating the analyte (10)
with one each first (20) and second probe (22) exhibiting an
affinity to the analyte (10), wherein the affinity of the
10 first probe (20) is caused by a specific affinity to at least
one first binding site (12) of the analyte (10) and the
incubating takes place under conditions under which the first
(20) and the second probe (22) bind to the analyte (10),

15 b) Labeling the first probe (20) with at least one electro-
chemically, specifically provable first marker (24), at least
when it is not already electro-chemically, specifically
provable,

20 c) Labeling the second probe (22) with at least one
electro-chemically, specifically provable second marker (26),
at least when it is not already electro-chemically,
specifically provable,

25 d) Abstracting the first (20) and second probe (22) bound
to the analyte (10),

e) Detection of a first electro-chemical signal (Si1)
caused by the abstracted first probe (20) or the first marker
30 (24) and a second electro-chemical signal (Si2) caused by the
abstracted second probe (22) or the second marker (26) and

f) Proof, quantification and/or characterization of the
analyte (10) by means of a ratio between the first (Si1) and
35 the second signal (Si2).

2. Method for the proof, quantification and/or characterization of an analyte (10) contained in a first fluid consisting of the following steps:

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- a) Bringing into contact and incubating the analyte (10) with one each first probe (20) exhibiting an affinity to the analyte (10), wherein the affinity of the first probe (20) is caused by a specific affinity to at least one first binding site (12) of the analyte (10) and incubating takes place under conditions under which the first probe (20) binds to the analyte (10),
- b) Labeling the first probe (20) with at least one electro-chemically, specifically provable first marker (24), at least when it is not already electro-chemically, specifically provable,
- c) Labeling the analyte (10) with at least one electro-chemically, specifically provable second marker (26), at least when the analyte (10) is not already electro-chemically, specifically provable,
- d) Abstracting the analyte (10) bound by the first probe (20) and the first probe (20) bound by the analyte (10),
- e) Detection of a first electro-chemical signal (Si1) caused by the abstracted first probe (20) or the first marker (24) and a second electro-chemical signal (Si2) caused by the abstracted analyte (10) or the second marker (26) and
- f) Proof, quantification and/or characterization of the analyte (10) by means of a ratio between the first (Si1) and the second signal (Si2).

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3. Method according to claim 1 or 2, wherein an electrode (27) used for the detection is not brought into contact with the first fluid.

5 4. Method according to one of the preceding claims, wherein the analyte (10) is separated from the first fluid before step lit. e, in particular before step lit. a and is transferred to a second fluid.

10 5. Method according to one of the preceding claims, wherein the analyte (10) is bound in particular specifically by a catcher molecule (pT, (T)_n).

15 6. Method according to one of the preceding claims, wherein the catcher molecule (pT, (T)_n) is immobilized on a first (16) or second surface.

20 7. Method according to one of the preceding claims, wherein the catcher molecule (pT, (T)_n) is a nucleic acid, an analogue of a nucleic acid, particularly a peptide nucleic acid (PNA), an antibody or a receptor.

25 8. Method according to one of the preceding claims, wherein the catcher molecule (pT, (T)_n) has an affinity molecule, particularly streptavidin, avidin or biotin, or a biotinylated oligonucleotide.

30 9. Method according to one of the preceding claims, wherein the first (20) or second probe (22) or the analyte (10) contains an affinity molecule, in particular biotin, avidin or streptavidin.

35 10. Method according to one of the preceding claims, wherein the analyte (10) is a nucleic acid having in particular a poly-T-end (pT, (T)_n) or a poly-A-end (pA, (A)_n).

11. Method according to one of the preceding claims, wherein the characterization takes place by determination of the length of the nucleic acid.

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12. Method according to one of the preceding claims, wherein the analyte (10) is amplified before or during step lit. a by means of a nucleic acid amplification reaction, in particular a PCR.

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13. Method according to one of the preceding claims, wherein the analyte (10) is immobilized before step lit. d, in particular before step lit. a, on the first (16) or second surface.

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14. Method according to one of the preceding claims, wherein the first surface (16) is the surface of a particle (18), particularly a superparamagnetic one.

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15. Method according to claim 14, wherein the particle (18) has a diameter of 10 nm to 100 μm , in particular 1 - 10 μm .

16. Method according to one of the preceding claims, wherein the second surface is an electrode (27) used for detection, in particular containing an electrically conductive plastic or an electrically conductive polymer, mercury, amalgam, gold, platinum, carbon or indium tin oxide.

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17. Method according to one of the preceding claims, wherein the second probe (22) exhibits a specific affinity to at least one specific second binding site (14) of the analyte (10).

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18. Method according to the preceding claim, wherein the analyte (10) exhibits a known number of first binding sites (12) and an unknown number of second binding sites (14).

5 19. Method according to one of the preceding claims, wherein the analyte (10) is a DNA fragment which exhibits repetitive sequences as the consequence of a triplet expansion disease such as fragile X syndrome, Huntington's disease, bulbar muscular atrophy, type I spinocerebral ataxia, myotonic
10 dystrophy or Friedreich's ataxia.

20. Method according to one of the preceding claims, wherein the first probe (20) is released from the analyte (10) and/or the analyte (10) is released from the first (16) or second
15 surface between step lit. d and step lit. e, in particular via heat denaturation, chemical denaturation, enzymatic digestion or chemical breakdown.

21. Method according to one of the preceding claims, wherein
20 the first (20) and the second probe (22) are released separately from one another from the analyte (10) or the first probe (20) from the analyte (10) and the analyte (10) from the first (16) or second surface and then detected.

25 22. Method according to one of the preceding claims, wherein the first marker (24) and/or the second marker (26) are released, in particular separately from one another, from the first (20) and/or second probe (22) or the analyte (10) and then detected.

30 23. Method according to one of the preceding claims, wherein the first (24) and/or the second marker (26) are released by enzymatic digestion or chemical breakdown.

24. Method according to one of the preceding claims, wherein the first (20) and/or second probe (22) is a nucleic acid or an analogue of a nucleic acid, in particular a peptide nucleic acid (PNA).

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25. Method according to one of the preceding claims, wherein the first (20) and/or second probe (22) binds sequence-specifically to the analyte (10), in particular via hybridization.

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26. Method according to one of the preceding claims, wherein the first (Si1) or second signal (Si2) is each caused by a catalytic hydrogen release caused by the first (24) or second marker (26).

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27. Method according to one of the preceding claims, wherein the first (24) and/or the second marker (26) can be reversibly reduced or oxidized.

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28. Method according to one of the preceding claims, wherein the first (24) or second marker (26) has an osmium complex, a nano gold particle, a cysteine, ferrocenyle, daunomycin, benzoquinone, naphthoquinone, anthraquinone or p-aminophenol group or a dye, in particular indophenol, thiazine or

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phenazine.

29. Method according to one of the preceding claims, wherein the first (20) or second probe (22) is labeled by several markers (24, 26).

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30. Method according to one of the preceding claims, wherein the first (20) or second probe (22) has a linear primary structure on whose one end the marker (24, 26) is located.

31. Method according to one of the preceding claims, wherein the detection of the first (Si1) and the second signal (Si2) takes place on the same electrode and/or via the same electro-chemical method of proof.

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32. Method according to one of the preceding claims, wherein the detection takes place by means of cathodic stripping voltammetry (CSV), squarewave voltammetry, cyclic voltammetry or chronopotentiometry.

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33. Method according to one of the preceding claims, wherein the detection takes place by means of a reversible redox process or a catalytic hydrogen development.